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Limbal Stem Cell Allografts and Corneal Transplant in a Patient with Severe Corneal Melting and Perforation due to Thermokeratoplasty and Cross-Linking Treatment Burn

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Key Words

Stem cells · Corneal melting · Thermokeratoplasty · Thermal burn

Abstract

Purpose: To report corneal stem cell allografts in a patient with a persistent epithelial defect as well as corneal melting and perforation due to severe ultraviolet light burn and thermokeratoplasty treatment for keratoconus.

Methods: A 21-year-old female patient with corneal melting, perforation and a persistent epithelial defect in her left eye secondary to iatrogenic treatment for keratoconus, thermokeratoplasty and cross-linking was treated with penetrating keratoplasty, using a 9.0-mm diameter corneal graft and limbal stem cell allograft implants. At the end of the procedure, subtenonian injections of a combination of bevacizumab and triamcinolone were given.

Results: The patient had a favorable outcome 48 h after surgery, with an improvement of symptoms and a complete corneal healing. By the third week after surgery, she had a best-corrected visual acuity of 20/60 and a clear corneal graft, which remained stable for the 9 months of follow-up.

Conclusions: Treatment with limbal stem cell allografts and penetrating keratoplasty in a female patient with a large corneal defect and melting in her left eye was effective. Larger studies are warranted to explore the real impact of this procedure.

Introduction

The ocular surface consists of two distinct types of epithelial cells, constituting the conjunctival and the corneal epithelium. Other regions with special kinds of epithelial cells are the limbal palisades of Vogt and the interpalisade ridges that are believed to be repositories of stem cells.

Several factors present in the microenvironment of the limbal region are considered important in maintaining the stemness of stem cells.

In some ocular surface diseases like thermal or alkali burns, stem cell transplantation has become one of the mainstays of management to improve the prognosis of patients, especially if they eventually undergo penetrating keratoplasty (PK). This is thought to improve the survival of the corneal graft.

Case Presentation

A 21-year-old female patient with a previous diagnosis of keratoconus had a corneal perforation of her left eye for 3 weeks. She had previously undergone a combined procedure of phacorefractive surgery with intraocular lens implantation, thermokeratoplasty and cross-linking treatment. At presentation, her left eye's visual acuity was light perception with a severe pain, conjunctival injection as well as corneal neovascularization, a large epithelial defect, a positive Seidel test, presence of a paracentral cyanoacrylate patch and multiple nylon sutures in the cornea ([fig. 1](#)).

Surgical Technique

Under local anesthesia, a lid speculum was sized and positioned to minimize pressure on the eye either from the speculum itself or indirectly from the lids. The first part of the procedure consisted of gently removing the nylon sutures, cyanoacrylate patches and two bandage contact lenses from the previously failed surgical procedure. The optical axis was then marked using the center of the pupil. The host cornea was marked by applying brief gentle pressure with a hand-held trephine blade. The donor cornea was then trephined with the endothelial side facing up, using a 9.0-mm donor trephine blade. The donor tissue was kept submerged in storage medium while the host corneal bed was prepared.

The host cornea was trephined using a hand-held disposable 8.5-mm trephine blade held perpendicular to the cornea. Viscoelastic material was placed through a small limbal paracentesis site at the 9 o'clock position, using a 15-degree blade prior to the full-thickness trephination to allow some cushioning upon entry to the anterior chamber. Minimal pressure was exerted against the cornea as the trephine was progressively being rotated, allowing its sharp edges to gently penetrate into the pre-Descemet's membrane or until the anterior chamber was entered.

Recipient site preparation was completed with beveled corneal scissors, aided by maintaining host button alignment with a fine-toothed forceps 45–90° away. Remaining tags of tissue were trimmed with curved Vannas scissors. The anterior chamber was filled with a viscoelastic to help maintaining donor button orientation for accurate suture placement and to provide endothelial protection.

The donor corneal tissue was gently grasped with a fine-toothed forceps at the junction of the epithelium and stroma and transferred on to the recipient bed, resting on viscoelastic material. Afterwards, the first 10/0 nylon interrupted suture was placed at the 12 o'clock position. The donor cornea was grasped with a fine-toothed, double-pronged forceps at the epithelial-stromal junction, and the suture was passed directly under the forceps teeth through the donor and aligned host tissue. The suture was tied snugly, using an initial triple loop followed by two additional single loops. Additional viscoelastic was placed in the anterior chamber to help maintain proper graft orientation and anterior chamber depth. The second suture was placed 180° away at the 6 o'clock position.

Afterwards, the suture was tied, the anterior chamber was reformed and tissue alignment checked once again. The 3 o'clock suture was also placed and tied, followed by the 9 o'clock suture. Twelve

additional radial interrupted 10/0 nylon sutures were placed snugly to ensure adequate tissue apposition, but not tightly. The anterior chamber was once again reformed with viscoelastic. The knots were buried on the donor side. The wound was checked for leaks with a cellulose sponge.

The second part of the procedure consisted of cutting two 2 × 3-mm limbal allografts from the donor tissue with Vannas scissors and placing under the tenon capsule of the host limbus in the superonasal and superotemporal quadrants.

Subtenonian triamcinolone (Softram; Grin Inc., Mexico City, Mexico) and bevacizumab (Avastin; Genentech Inc., South San Francisco, Calif., USA) injections were applied to reduce corneal neovascularization and possible rejection of the corneal graft. Chloramphenicol ointment and a patch were applied for 48 h.

Residual bullae developed 9 months after the allografts were implanted in the superior limbus ([fig. 2](#)). Forty-eight hours after the procedure, there was a complete epithelial healing. Three weeks after surgery, best-corrected visual acuity was 20/60 with a clear corneal graft that remained stable for the 9 months of follow-up. There was no residual corneal neovascularization either ([fig. 3](#)).

Discussion

The purpose of this report was to describe the combination of limbal stem cell allografts and PK for the treatment of severe thermal burn and corneal perforation.

Limbal stem cell auto- and allograft have long been used for the treatment of multiple ocular surface pathologies, especially for those secondary to alkali burns where there is usually a severe epithelial insufficiency.

In their review, Ramaesh and Dhillon [1] state that the management of severe ocular surface disease due to limbal stem cell deficiency has changed dramatically in recent years. The concept of limbal stem cells as the source of corneal epithelium revolutionized the therapeutic approach of ocular surface reconstruction. Deficiency of limbal stem cells results in blinding ocular surface diseases.

Grafting viable limbal tissue, from either a fellow healthy eye or a donor eye, with the resident stem cell population may replenish limbal stem cells and can restore the corneal surface to normality. Transplanting limbal tissue can be achieved through a variety of procedures that include cadaveric keratolimbal allograft, live or living-related conjunctival-limbal allograft and limbal autograft [2].

Advances in tissue engineering techniques have offered a viable alternative to overcome the scarceness of limbal tissue available for transplantation. Epithelial stem cells harvested from a small limbal biopsy can be expanded in vitro on a suitable carrier and then transplanted to the diseased cornea to restore the corneal surface successfully [3]. There are many reports using allograft of limbal stem cells to treat limbal insufficiency.

Holland [2] defines epithelial transplantation for ocular surface diseases as one of the following procedures: conjunctival autograft, conjunctival allograft, conjunctival-limbal autograft, cadaveric conjunctival-limbal allograft, living-related conjunctival-limbal allograft or keratolimbal allograft.

Their evaluation of their keratolimbal allograft patients revealed that 18 of 25 eyes (72%) developed a stable ocular surface. Fifteen eyes (60%) demonstrated a significant improvement in visual acuity. Persistent epithelial defects and symblephara were

successfully managed with this procedure. Six of 13 (46%) subsequent keratoplasties were successful.

Patients with limbal deficiency due to Stevens-Johnson syndrome had a significantly worse outcome. Patients with preoperative conjunctival keratinization also had a worse outcome [4].

Compared with our report, the better final visual and anatomic outcome might be because although severe melting and large epithelial defects were present, thermal burns due to thermokeratoplasty and cross-linking treatment are less associated with severe epithelial insufficiency.

In his study, Miri et al. [5] used and studied the survival of the limbal allograft, was seen in 22 eyes (82%). Within 3 months, the surface failed in 4 eyes, and after 43 months, it failed in the fifth patient. In our case report, we describe the outcome at only 9 months after the procedure, but it is likely that a long survival of the corneal graft may be achieved.

Mitra [6] described another successful outcome using limbal allograft in a corneal chemical burn. In this case, the prognosis tends to be worse and not every patient has a long stem cell survival.

To the best of our knowledge, this is the first report that uses epithelial limbal cell allografts to enhance the prognosis of PK for large corneal perforation after ultraviolet light treatment in a cross-linking procedure, and for corneal burns as well as persistent epithelial defects due to thermokeratoplasty.

One of the drawbacks of this report is that it only describes 1 single case, which does not conclusively reflect the exact role of this treatment modality for persistent epithelial defects due to ultraviolet light and thermokeratoplasty burns. Larger comparative series are necessary to elucidate the real effectiveness of this surgical approach.

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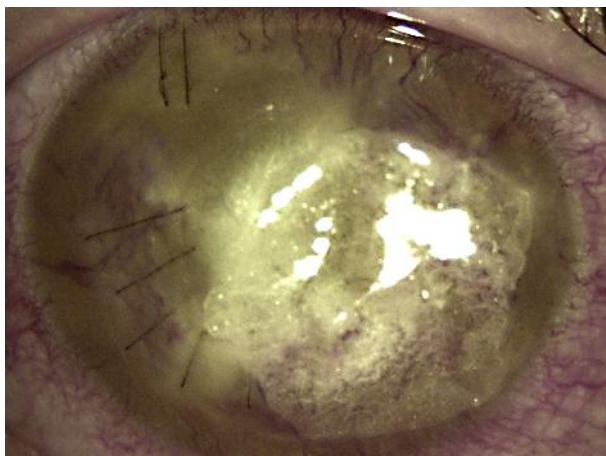


Fig. 1. Conjunctival injection, corneal neovascularization, a large epithelial defect, presence of a paracentral cyanoacrylate patch and multiple nylon sutures in the patient's left eye cornea at presentation.

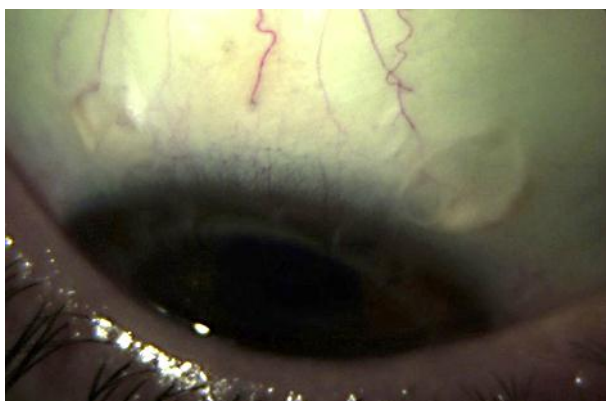


Fig. 2. Residual bullae 9 months after surgery where the allografts were implanted in the superonasal and superotemporal limbus of the patient's left eye.

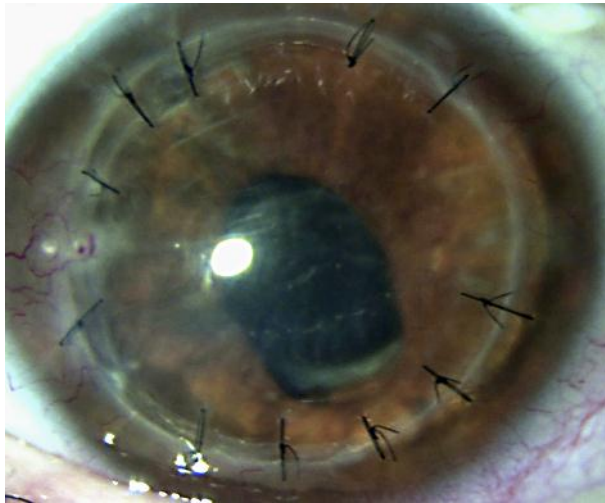


Fig. 3. Clinical picture 9 months after surgery, showing a clear corneal graft and a regressed neovascularization.

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